

Page 11, third line of first partial paragraph, delete "paralell" and insert therefor --parallel--; ninth line of first full paragraph, delete "warrented" and insert therfor --warranted--; first line of second full paragraph, delete "warrantied" and insert therefor --warranted--; seventh line of last paragraph, delete "our" and insert therfor --or--.

### In the Claims

Please cancel claims 1-21 without prejudice of disclaimer of the subject matter therein. Please add claims 22-79 as follows:

22. (New) A method for detecting a specific target cell in a cell suspension of a mixed cell population, in a fluid system containing a mixed cell population, or in a single cell suspension prepared from a solid tissue, with the exception of normal and malignant hematopoietic cells in blood and bone marrow, the method comprising the steps of:

a. coating paramagnetic particles or beads with an antibody or antibody fragment directed against a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture;

b. mixing the coated paramagnetic particles or beads with the cell suspension containing the target-cells;

c. incubating the mixture under gentle rotation;

d. examining the target-cells after incubation; and

e. counting the target-cells after incubation.

*Sub*  
~~23. (New) The method of claim 22, wherein the paramagnetic particle or bead is coated with a murine or a human antibody or fragment thereof.~~

~~24. (New) The method of claim 22, wherein incubating lasts for 5-10 minutes to 2 hours.~~

~~25. (New) The method of claim 24, wherein incubating lasts 30 minutes.~~

*D*  
~~26. (New) The method of claim 22, wherein incubating is at a temperature between 0°C and 25°C.~~

~~27. (New) The method of claim 26, wherein incubating is at a temperature of about 4°C.~~

*Sub*  
~~28. (New) The method of claim 22, wherein when the target cell population is contained in blood or bone marrow aspirates, the method further comprises the step of:  
pre-incubating the antibody-coated paramagnetic particle and the cell suspension with mild detergent.~~

~~29. (New) The method of claim 28, wherein the preincubating comprises as detergent Tween 20™ at a concentration less than 0.1% and the preincubation lasts 30 minutes at 4°C.~~

~~30. (New) The method of claim 22, wherein when the density of target-cells is low, or when the ratio of target cell/total cells in the cell mixture is low ( $\leq 1\%$ ), the method further comprises the step of:~~

subjecting the incubated paramagnetic particle-antibody-cell mixture to a magnetic field.

31. (New) The method of claim 30, wherein the particle-target-cell complexes are stained.

32. (New) The method of claim 22, wherein the step of examining, the step of counting, or both steps comprise using a microscope or a cell or particle counting device.

33. (New) The method of claim 22, further comprising the steps of:

isolating the target-cells by exposing the complex of cells and paramagnetic particles to a magnetic field to magnetically aggregate the cells;

subjecting the magnetically aggregated cells to further biological, biochemical, and immunological examination.

34. (New) The method of claim 22, wherein the antibody or fragment thereof is directed against an antigen or a receptor in a cell with abnormal developmental patterns

35. (New) The method of claim 34, wherein the cell is a primary or a metastatic cancer cell.

36. (New) The method of claim 22, wherein the antibody or fragment is of IgG isotype, a F(ab')<sub>2</sub> fragment, a F(ab) fragment, IgM, or a fragment of IgM.

37. (New) The method of claim 22, wherein the cell suspension or population comprises mammalian tissue, a pleural effusion, a peritoneal effusion, a body fluid, or a solid tumor in a normal tissue or organ.

38. (New) The method of claim 37, wherein the mammalian tissue comprises human bone marrow or human peripheral blood; the body fluid comprises urine, cerebrospinal fluid, semen, or lymph; or the normal tissue or organ comprises liver, lymph node, spleen, lung, pancreas, bone, central nervous system, prostate gland, skin, or mucous membranes.

39. (New) The method of claim 22, wherein the antibody or antibody fragment is directed against fibronectin receptor,  $\beta$ -integrin, vitronectin receptor,  $\alpha\beta$ 3-integrin, P-selectin, GMP-140, CD44-variants, N-CAM, E-cadherin, Le<sup>y</sup>, CEA, EGF receptor, c-erbB-2, HER2, transferin receptor, TNF-receptor, high molecular weight antigen (HMW 250,000), p95-100, TP-1 and TP-3 epitope,, Mv 200kD, Mv160kD, MOC-31 epitope, cluster 2 epithelial antigen, MUC-1 antigen, DF3-epitope, gp290kD, prostate high molecular antigen (Mv>400kD), TAG 72, bladder carcinoma antigen (Cancer Res. 49, 6720, 1989), Mv 48kD colorectal carcinoma antigen, lung carcinoma antigen Mv 350-420kD, Mel-14 epitope,  $\beta_2$ -microglobulin, Apo-1 epitope, or pan-human cell antigen.

40. (New) The method of claim 22, wherein the antibody or antibody fragment is directed against a growth factor

receptor and or an oncogene product expressed on the membrane of a malignant cell

41. (New) The method of claim 40, wherein the antibody or antibody fragment is directed against an insulin receptor, an insulin-like receptor, or FGF.

42. (New) The method of claim 34, wherein the antibody or antibody fragment is directed against an adhesion membrane molecule or an MDR proteins in the abnormal cell.

43. (New) The method of claim 34, wherein the antibody or antibody fragment is directed against breast, ovarian or lung carcinoma cells; melanoma, sarcoma, glioblastoma or cancer cells of the gastrointestinal tract; melanoma, sarcoma, glioblastoma or cancer cells of the genitourinary tract; or melanoma, sarcoma, glioblastoma or cancer cells of the reticuloendothelial system.

44. (New) The method of claim 34, wherein the antibody or antibody fragment is directed against cells associated with a non-neoplastic disease.

45. (New) The method of claim 44, wherein the non-antineoplastic disease affects a cardiovascular cell, a neurological cell, a pulmonary cell, an autoimmune cell, a gastrointestinal cell, a genitourinary cell, or a recticuloendothelial cell.

Sub  
E4  
46. (New) A kit for performing the method of claim 22, the kit comprising:

a. a specific antibody or antibody fragment directed to an antigen on a target-cell, which antibody or fragment is effective for coating a paramagnetic particle or bead without removing its antigen-binding ability;

b. a paramagnetic particle or bead; and

c. another specific antibody or antibody fragment directed against an antigen or a receptor within or on the target cell;

wherein said another antibody or antibody fragment is conjugated to biotin or to an enzyme; or wherein said another antibody or antibody fragment is bound to a non-paramagnetic particle with a specific color or with a bound enzyme.

47. (New) The kit of claim 46, wherein the enzyme is peroxidase or alkaline phosphatase.

48. (New) A method for detecting a specific target cell in a cell suspension of a mixed cell population, in a fluid system containing a mixed cell population, or in a single cell suspension prepared from a solid tissue, with the exception of normal and malignant hematopoietic cells in blood and bone marrow, the method comprising the steps of:

a. pre-coating paramagnetic particles or beads with an antibody directed against an Fc-portion of an antibody or antibody fragment directed against a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture;

- Sub  
E6
- b. forming a complex comprising the pre-coated paramagnetic particles, the antibody or antibody fragment directed against a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture, and the target-cell;
  - c. examining the target-cells in the complex; and
  - d. counting the target-cells in the complex.

49. (New) The method of claim 48, wherein the step of forming a complex comprises:

coating the pre-coated paramagnetic particles or beads with an antibody or antibody fragment directed against a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture;

mixing the coated, precoated paramagnetic particles or beads with the cell suspension containing target-cells; and  
incubating the mixture under gentle rotation.

50. (New) The method of claim 49, wherein incubating lasts for 5-10 minutes to 2 hours.

51. (New) The method of claim 49, wherein incubating lasts 30 minutes.

52. (New) The method of claim 49, wherein incubating is at a temperature between 0°C and 25°C.

53. (New) The method of claim 49, wherein incubating is at a temperature of about 4°C.

54. (New) The method of claim 48, wherein the step of forming a complex comprises:

mixing antibodies directed against a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture with the cell suspension containing the target cells;

incubating the mixture under gentle rotation;

adding the pre-coated paramagnetic particles or beads to the incubating mixture; and

continuing the incubation.

55. (New) The method of claim 54, wherein incubating lasts for 5-10 minutes to 2 hours.

56. (New) The method of claim 54, wherein incubating lasts 30 minutes.

57. (New) The method of claim 54, wherein incubating is at a temperature between 0°C and 25°C.

58. (New) The method of claim 54, wherein incubating is at a temperature of about 4°C.

59. (New) The method of claim 48, wherein the antibody or antibody fragment directed against a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture is a murine or a human antibody or fragment thereof.



60. (New) The method of claim 48, wherein when the target cell population is contained in blood or bone marrow aspirates, the method further comprises the step of:

pre-incubating the antibody-coated paramagnetic particle and the cell suspension with mild detergent.

61. (New) The method of claim 48, wherein the preincubating comprises as detergent Tween 20™ at a concentration less than 0.1% and the preincubation lasts 30 minutes at 4°C.

62. (New) The method of claim 48, wherein when the density of target-cells is low, or when the ratio of target cell/total cells in the cell mixture is low ( $\leq 1\%$ ), the method further comprises the step of subjecting the complex to a magnetic field.

63. (New) The method of claim 62, wherein the particle-target-cell complexes are stained.

64. (New) The method of claim 48, wherein the step of examining, the step of counting, or both steps comprise using a microscope or a cell or particle counting device.

65. (New) The method of claim 48, further comprising the steps of:  
isolating the target-cells by exposing the complex of cells and paramagnetic particles to a magnetic field to magnetically aggregate the cells;

subjecting the magnetically aggregated cells to further biological, biochemical, and immunological examination.

66. (New) The method of claim 48, wherein the antibody or fragment thereof is directed against an antigen or a receptors in a cells with abnormal developmental patterns

67. (New) The method of claim 66, wherein the cell is a primary or metastatic cancer cell.

68. (New) The method of claim 48, wherein the antibody or fragment is of IgG isotype, a F(ab')<sub>2</sub> fragment, a F(ab) fragment, IgM, or a fragment of IgM.

69. (New) The method of claim 48, wherein the cell suspension or population comprises mammalian tissue, a pleural effusion, a peritoneal effusion, a body fluid, or a solid tumor in a normal tissue or organ.

70. (New) The method of claim 69, wherein the mammalian tissue comprises human bone marrow or human peripheral blood; the body fluid comprises urine, cerebrospinal fluid, semen, or lymph; or the normal tissue or organ comprises liver, lymph node, spleen, lung, pancreas, bone, central nervous system, prostate gland, skin, or mucous membranes.

71. (New) The method of claim 48, wherein the antibody or antibody fragment is directed against fibronectin receptor,  $\beta$ -integrin, vitronectin receptor,  $\alpha\beta$ 3-integrin, P-

Sub  
E11  
selectin, GMP-140, CD44-variants, N-CAM, E-cadherin, Le<sup>y</sup>, CEA, EGF receptor, c-erbB-2, HER2, transferin receptor, TNF-receptor, high molecular weight antigen (HMW 250,000), p95-100, TP-1 and TP-3 epitope,, Mv 200kD, Mv160kD, MOC-31 epitope, cluster 2 epithelial antigen, MUC-1 antigen, DF3-epitope, gp290kD, prostate high molecular antigen (Mv>400kD), TAG 72, bladder carcinoma antigen (Cancer Res. 49, 6720, 1989), Mv 48kD colorectal carcinoma antigen, lung carcinoma antigen Mv 350-420kD, Mel-14 epitope,  $\beta_2$ -microglobulin, Apo-1 epitope, or pan-human cell antigen.

D  
X  
72. (New) The method of claim 48, wherein the antibody or antibody fragment is directed against a growth factor receptor and or an oncogene product expressed on the membrane of a malignant cell

73. (New) The method of claim 72, wherein the antibody or antibody fragment is directed against an insulin receptor, an insulin-like receptor, or FGF.

74. (New) The method of claim 66, wherein the antibody or antibody fragment is directed against an adhesion membrane molecule or an MDR proteins in the abnormal cell.

Sub  
E12  
75. (New) The method of claim 66, wherein the antibody or antibody fragment is directed against breast, ovarian or lung carcinoma cells; melanoma, sarcoma, glioblastoma or cancer cells of the gastrointestinal tract; melanoma, sarcoma, glioblastoma or cancer cells of the

*Jul ER*  
genitourinary tract; or melanoma, sarcoma, glioblastoma or cancer cells of the reticuloendothelial system.

76. (New) The method of claim 66, wherein the antibody or antibody fragment is directed against cells associated with a non-neoplastic disease.

77. (New) The method of claim 76, wherein the non-antineoplastic disease affects a cardiovascular cell, a neurological cell, a pulmonary cell, an autoimmune cell, a gastrointestinal cell, a genitourinary cell, or a reticuloendothelial cell.

*D*  
78. (New) A kit for performing the method of claim 48, the kit comprising:

- AB*
- a. a first specific antibody or antibody fragment directed to a membrane structure specifically expressed on the target-cell and not on a non-target-cell in the cell mixture;
  - b. a second antibody or antibody fragment directed to an Fc-portion of the first antibody, which second antibody or fragment is effective for coating a paramagnetic particle or bead without removing its antigen-binding ability;
  - c. a paramagnetic particle or bead; and
  - d. a third specific antibody or antibody fragment directed against an antigen or a receptor within or on the target cell;

wherein said third antibody or antibody fragment is conjugated to biotin or to an enzyme; or wherein said third antibody or antibody fragment is bound to a non-paramagnetic particle with a specific color or with a bound enzyme.